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RESEARCH ARTICLE

PROXIMATE ANALYSIS FOR BIOACTIVE COMPOUNDS IN LEAF EXTRACT OF BAPHIA NITIDA Lodd.

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ABSTRACT

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Key words:

Baphia nitida, Bioactive compounds, Screening, and biological activity. Preliminary screening was carried out on *Baphia nitida* to establish the presence of certain bioactive compounds. Alcohol extract of fresh leaves of *B. nitida* was obtained and several identification tests, each specific for detecting the presence of a particular chemical compound were carried out, using standard techniques, on the alcohol extract. The chemical compounds tested for include alkaloids, glycosides, tannins, saponnins, flavonoids, reducing compounds, polyphenols, phlobatinnins and anthraquinones. The results showed that polyphenols showed heavy presence and alkaloids, glycosides, tannins, saponins and reducing compounds were present in light amounts. The findings of this test present a rough estimation of the biological (pharmacological) activities of leaf extract of *B. nitida*. Practically, the actual biological (pharmacological) activities of the individual compounds can only be determined by isolating and testing of these compounds on experimental animals

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INTRODUCTION

Nature has been a source of medicinal agents for thousands of years and generally produces many secondary metabolites which constitute important leads for the development of new environmentally friendly microbicides, pesticides, herbicides, and many pharmaceutical drugs (Bobbarala et al., 2009). The exclusive use of herbal remedies to treat and manage ailments had served from the onset as the most important therapeutic approach available to man. However, the decline from its use due to introduction of modern synthetic medicine, started at about the beginning of the 20th century up to the 1970s (Wills et al., 2000). The extraction of bioactive agents from plants is one of the most intensive areas of natural product research today, yet the field is far from exhausted. Many different approaches may be followed in the screening of plants for products of pharmacological and chemical interest. Basic phytochemical screening consists of performing simple chemical test to detect the presence of alkaloids, tannins, saponins, digitalis glycosides, etc in plant extracts (Sofowora, 1982). The prospective investigator should consider two fundamental issues. Why it is necessary to attempt to obtain bioactive agents from plants, and the objectives of the exercises.

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Department of Botany, Nnamdi Azikiwe University, P.M.B. 5025, Awka, Nigeria *Baphia nitida* is a dicotyledonous angiosperm belonging to the family papilionaceae. The plant is a shrub growing to about 3 meters on the plains and about 10 meters in the forests. It possesses very smooth branchlets with 1-foliate leaves, petioles about 13-25 mm long. The flowers occur in axillary fascicles or solitary with glabrous ovary. They are conspicuously white with yellow center. The fruits are oblanceolate, up to 15 cm long and often glabrous. *B. nitida* is important economically and often cultivated for its red dye, which is soluble in alkali. The plant is used in making dye. The red dye is extracted from the bark and the heartwood and is found to contain a chemical santalic acid (santalin).

Dye wood or Cam wood is also used in making tribal marks and tattoos among certain tribes in West Africa sub region, especially in Ivory Coast, Benin Republic and Togo. The fragrance of the flowers is exploited in making perfume in the cosmetic industry. In Nigeria, the use of natural products for treatment cannot be overlooked due to the large number of country's population and the inadequacy of our health care system (Sade, 2002). Investigations of folk medicine have resulted in the discovery of some phytochemicals responsible for the therapeutic actions of plants on living organisms. For example, the leaves of the species are used in folk medicine for the treatment of inflamed infected umbilical cord in Nigeria (Contus, 2012). Against this general background, the researchers have sought to carry out preliminary phytochemical analysis to establish the presence of bioactive compounds in the leaves of *B. nitida*.

MATERIALS AND METHODS

Study Area and Materials

The disease-free plant material used for the analysis was collected from a forest in Ikot Effanga Mkpa area in Calabar Municipality, Cross River State, Nigeria. A branch from the species was cut with a matchet and conveyed in a dry polythene bag to the Department of Botany, University of Calabar, Nigeria where the species was properly identified as *Baphia nitida*. The leaves of the specimen, *Baphia nitida* were washed with distilled water to remove sands and other particles and then dried in the sun until they were properly dried. The dried samples were then pulverized using electric blender to obtain fine powder. The pulverized sample with characteristic dark-green colour was preserved and stored in an air-tight container for phytochemical analysis.

Chemicals and Reagents

All chemicals and reagents used were of analytical standards. They include:

- 98% absolute alcohol
- 1% aqueous hydrochloric acid
- Mayer's reagent
- Dragendorff''s reagent
- Chloroform
- Sulphuric acid
- 1% ferric chloride
- Aluminium metal
- Fehling's solution
- 1% ammonium solution
- Acetyl anhydride
- Concentrated sulphuric acid

Preparation of Alcohol Extract of Baphia nitida

Twenty grammes (20g) of well ground powdered specimen of *B. nitida* was sbjected to extraction with 100ml of 98% absolute ethanol for 40mins in a continuous extraction (soxhlet) apparatus. The alcoholic extract was filtered through a cheese cloth and concentrated to 50ml. The extract was cooled and preserved for further analysis.

Phytochemical Analysis

The following tests were carried out on the extract:

Identification of alkaloids

Using Mayer's and Dragendorff test (Sofowora, 1984)

2 cm2 of plant extract was stirred with 5cm2 of 1% aqeous HCL on a water bath. 1 cm3 of the filtrate was treated with a few drops of Mayer's reagent and a second 1cm3 with Dragendorff reagent. Turbidity or precipitates either of these reagents was taken as preliminary evidence for the presence of alkaloids.

Identificatiuon of Glycosides

Using Salkowsk Test (Sofowora, 1984)

2cm3 of plant extract was dissolved in 2cm3 of chloroform. Sulphuric acid was carefully added to form a lower layer. A reddish-brown colouration at the interface indicates the presence of glycosides

Identification of Tannins

Using Ferric Chloride Test (Trease and Evans. 1978)

In this test, 2cm3oftract with 10cm3 of distilled water and heated in the water bath. 1ml of 1% Fecl3 was added to the filterate. Blue-black, green or blue-green precipitate indicates presence of tannins.

Identification of Flavonoids

Using Aluminium Chloride Test

Here, 2cm3 of the extract was added to a few pieces of aluminium metal and conc. HCL added. The formation of orange, red, crimson or magneta indicates the presence of flavonoids.

Identification of Reducing Compounds

Using Fehling's Test

In this test, 2cm3 of the plant extract was put in a test tube and 5cm3 of the fehling solution added to it and heated in the water bath for 5mins. The formation of brick-red precipitate or colouration indicates the presence of reducing compounds.

Identification of Polyphenol

Potassium Ferrocyanide Test

Here, 2cm3 of the plant extract was treated with 5cm3 of distilled water and heated for 30mins in a water bath. 1ml of 1% Fecl3 added to the solution. The mixture was filtered. The formation of green-blue colouration indicates the presence of polyphenol.

Identification of Phlobatannins

Using Aqueous Hydrochloric acid Test (Trease and Evans, 1989)

In this test, 2cm3 of plant extract was boiled with 5cm3 of 1% aqueous solution of HCL. Deposition of red colour of or precipitate indicates the presence of phlobatannins

Identification of Anthraquinones

Using Borntrager;s Test (Trease and Evans, 1989)

Here, 2cm3 of the plant extract was shaken with 10ml of benzene. This was filtered and 5ml of 105 NH3 added. The mixture was shaken and the presence of pink-red or violet colouration in ammoniacal (lower) p[hase indicates the presence of free anthraquinones.

RESULTS AND DISCUSSION

The results of the phytochemical screening of alcohol extract of *B. nitida* are presented in Table1.

 Table 1. Occurrence of Bioactive Compounds in Alcohol Extract of Baphia nitida

Bioactive Constituent	B. nitida
Alkaloids	+
Glycosides	+
Tannins	+
Saponins	+
Flavonoids	-
Reducing compounds	+
Polyphenols	++
Phlobatannins	-
Anthraquinones	-

Key

- + indicates presence in slight amounts
- ++ indicates presence in heavy amounts
- indicates nil presence

The results in Table 1 shows that the alcohol extract of the leaves of *Baphia nitida* ains heavy amounts of polyphenol while alkaloids, glycosides, tannins, saponins and reducing compounds were present in light amounts. However, the extracts did not contain flavonoids, phlobatannins, and anthraquininones.

The presence of the above mentioned compounds is indicative of the fact that *Baphia nitida* is valuable for use as a medium for alleviation of certain diseases. Sofowora (1983) reported that the medicinal properties of plants are related to the phytochemocal constituents present in them. It is therefore probable that this plant which contains important phytochemical compounds in its extract has potential for use against certain diseases. For example, phytochemicals such as saponins have anti-inflammatory effects (Vinha and Soares, 2012), haemolytic activity, and cholesterol binding properties (Nyarko and Addy, 1990). Glycosides are known to lower blood pressure (Marinkovic and Vitale, 2008) and tannins exhibit antioxidant, antimicrobial and antiviral effects (Sayyah and Hadida, 2004).

Most alkaloids are poisonous with marked pharmacological activities. Alkaloids have a more or less marked action on the Central Nervous System (CNS) and often, also in the peripheral nervous system (Trease and Evans, 1989). But this property is of therapeutic significance because when ingested they increase the secretion of digestive juices and so increase the appetite of the patient (Trease and Evans, 1989). Phytochemical compounds apart from their therapeutic properties can be used for industrial purposes. The term 'tannin' was first applied in 1976 to denote a range of plant polyphenols which possessed the property of precipitating proteins from aqueous media (Trease and Evans, 1778). This distinctive property has permitted their use for at least 2000 years for the conversion of raw animal hides to durable permeable leathers (Swain *et al.*, 1979).

Conclusion

The aim of this work was to focus attention on the potential of *Baphia nitida* to yield important components for drugs by examining the bioactive components present in alcohol extract of this plant. Presently, though *B. nitida* yields important dye substances, it has not played a significant role in treatment of diseases. This work has highlighted the important chemical components of *B. nitida* which when harnessed could play a useful role in the pharmaceutical industry. Therefore, it is concluded that *B. nitida* has potential to be used in the drug manufacturing industry. It is thus suggested that further research work should focus on extraction and separation of individual components and testing them on experimental animals to highlight their biological activities.

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